weeks following the thymectomy. They also showed an increased irritability of the cerebral cortex and peripheral nerves and that injections of soluble calcium salts brought the irritability back to normal. MacCallum and Voegtlin[31] and also Erdheim[32] have claimed a relation exists between the parathyroid glands and calcium metabolism.[33]

THE SERUM DIAGNOSIS OF PREGNANCY AND ITS UNDERLYING PRINCIPLES.

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In his test for pregnancy, Abderhalden has opened a field which promises perhaps more for scientific investigation and for ultimate practical usefulness than any other present-day line of study. He and his pupils have devoted a great deal of brilliant investigation to the work of animal ferments, and this test for pregnancy, introducing new laboratory methods of almost unlimited possibilities, is the result of these studies. I shall not discuss the great mass of experiments dealing with this subject the results of which have been reported by various investigators within the past two years. It is too early for a critical survey. The exacting details of the method and the principles underlying it must be more thoroughly understood by those who attempt to follow Abderhalden's work before any effort can be made to reconcile results. I shall confine myself to a discussion of these principles and to a brief consideration of the technique.[1]

All metabolic processes are in last analysis an expression of ferment activity. Each body cell is armed with ferments designed not only to protect it from foreign invasion, but which enable it to meet its requirements for energy, maintain its structural integrity, and perform its specific functions. The study of these cell ferments, overshadowing in importance everything else in physiology (and perhaps in pathology), is after all still in its infancy, for we know nothing of the exact nature of the chemical composition of ferments, and are forced to recognize them solely by the character of their work.

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[31] Jour. Exper. Med., 1909, ix, 118.

[32] Frankfurt Zeitsch. f. Path., 1911, vii, 178.

[33] For review of literature regarding the influence of internal secretory organs on mineral metabolism see Bayliss and Staring, Erg. d. Physiol., v, p. 664; Vincent, Erg. d. Physiol., ix, p. 145; Internal Secretion and Ductless Glands, 1912 (Arnold); Oppenheimer, Hand. d. Biochemie, 1910, 3, part 1: Biedl, Innere Sekretion, 1913.

[1] References to the voluminous literature upon animal ferments have been omitted. This may be found in complete form in Abderhalden's Abwehrfermente des tierischen Organismus. Julius Springer, Berlin.

On first thought we should expect to find the studies of ferments simplest among the unicellular bodies. We should expect their metabolic processes to move along simpler lines and to unfold themselves more readily to view. On further consideration, however, it is seen that the metabolic processes of unicellular organisms must necessarily be more complex and more difficult of access than is the case with those organisms in whose construction many kinds of cells are concerned; for with the former, absorption and excretion run parallel, and it is extremely difficult, in fact, impossible, to study in detail their manifold metabolic processes. As we go higher in the animal scale, we find specialized groups of cells, each monopolizing certain functions and gradually losing others. Such cell groups with their special functions we recognize as organs. From this viewpoint we can regard the animal organism as a cell community, each member of which, working for the good of the whole, has its definite duties to perform.

The first attempt at formation of a cell community which we call symbiosis, is seen in that mutual coöperation between unicellular bodies. Here the cells, though separate and distinct, bind themselves together because of a common interest. A little higher up the scale and we come upon the various cell groups of multicellular bodies. When one of these groups has formed an alimentary tract, it alone, as Abderhalden points out, stands in physiological relationship to the outer world. It alone determines for the organism the nature of its absorbed foodstuffs.

The prime object of digestion is to protect the body against the invasion of foreign substances, a recognition of which fact is necessary for a proper understanding of the principles upon which Abderhalden bases his work. No foodstuffs are absorbed directly. Neither proteids, carbohydrates, nor fats undergo immediate absorption, nor can they be reduced directly to their metabolic end-products. Before it is suitable for absorption, each must be broken down to its simplest building stones. This is accomplished by means of ferments produced by the intestinal epithelium and the extrinsic glands. From the mass of amino-acids and other split products thus set free, the organism chooses such building stones as are suitable for use. What immediately follows is shrouded in some doubt. Since amino-acids could not be found in the blood, it was formerly believed that they did not circulate as such, but were united during absorption to form new albumins. Recently, however, Van Slyke and Meyer have found that amino-acids do circulate as such, and in this form are carried to the body cells.

The liver holds up and rapidly destroys any excess of these absorbed bodies. The situation of this organ, between the intestinal epithelium and the body cells, where it can scrutinize and deal with all absorbed substances, is a fortunate one; for it not only aids in the elaboration of nutritive substances, but also serves as an important defence against foreign invasion. These building stones, perhaps loosely coupled to other substances, are taken up by the muscles and other tissues, and must undergo still further elaboration before they become integral parts of the cell.

Each variety of cell, exhibiting, as we are forced to assume, a structure peculiar to itself, must necessarily select from the materials offered it those building stones which are peculiarly suited to its needs and reject those which it cannot use. This is accomplished by means of cell ferments through digestive processes which are to a certain extent at least identical in nature with those of the alimentary tract. The blood plasma—the source from which each cell draws its building material—is for all cells the same. Consequently we should expect in each kind of cell not only its own specific ferments, but in addition, ferments which are common to all cells. In the study of this, the machinery of the cell, we shall find perhaps "the surest and most direct evidence that cell function is dependent upon cell structure." Not every split-product thus formed is ready for immediate absorption. At times it must be coupled to some other substance before a satisfactory building stone results. Then follows further coupling of all these manifold building stones until the characteristic complex cell structure is completed.

As an example of such reconstruction, "An architect is directed to construct from a given building which has served a certain specific purpose, and therefore is of special design, another building destined to entirely different uses. He must first demolish the former. From the building stones thus obtained he proceeds along entirely new lines. Many of these are ready for immediate use; others must be reshaped; while still others are wholly unfit. In exactly the same manner the animal organism deals with the complicated cell material coming to it as foodstuffs. First occurs a splitting into the simple building stones, and then beyond the intestinal wall reconstruction follows according to entirely new plans."

We are accustomed to view ferment activity in too schematic a manner. The conversion by the yeast cell of dextrose into alcohol and carbon dioxide is no doubt a much more complicated process than we ordinarily think, and is probably divided into many intermediary reactions. The meaning to the cell of each of these separate reactions will, as Abderhalden points out, constitute an interesting problem for future investigation, the solution of which will no doubt permit valuable insight into cell metabolism. The cell works step by step, liberating gradually the energy contained in its absorbed substances, and probably never at one stroke converting them to metabolic end-products.

Though every body cell conforms in structure to a certain general ground-plan common to all, those organ cells which have taken on special functions are distinguished by certain additional peculiarities of structure. We must assume for each variety of cell this specific plan of structure, else how can we explain its specific functions? Certain cells produce specific secretions, and these secretions influence solely cells of a given kind, leaving numberless others untouched. The secretion itself, the cell which produces it, and the cell which feels its influence, must each enjoy a plan of structure peculiar to itself.

With this conception of specific cell structure, and to a certain extent specific cell metabolism, we assume not only that each variety of cell within the human organism possesses its own peculiar build, but also that to a certain extent there is a variation in a given cell between two individuals of the same species. We also recognize that this cell presents still wider variations in structure when derived from animals of different species, and so on down the scale. This demands an infinite variety of structure, while the building stones at our disposal are comparatively few. The following may be quoted to show that our assumption is entirely reasonable: With three building stones we can build six different structures; with four, twenty-four; and with twenty building stones the number of possible structures is 2,432,902,008,176,640,000.

Recent experiments indicate that while the cell albumins of a given organ differ according to the animal from which the organ comes, they at the same time present certain points of similarity. To produce this similarity it is necessary only that there recur always in each of these albumins a given group which, because of certain stereochemical arrangement offers to the specific ferment suitable point for attack. This is important when we come to consider the availability for human experiment of albumins derived from organs of the lower animals.

A striking illustration of the specific adjustment of secretions to cells of a certain structure is cited by Abderhalden in the peculiar condition known as *hermaphroditimus verus*. These peculiar animals possess upon one side an ovary and upon the other a testis, and exhibit upon one-half of the body male and upon the other half female types of development, each stopping sharply at the midline. We must acknowledge that the secretions from each of these glands is brought in contact with all of the cells of both sides of the body; yet each brings to full development the secondary sexual characters peculiar to itself and limits them to its own body half. The secretion from the ovary finds suitable adjustment only in those body cells of the female side, and that of the testis influences only those cells built according to the male ground-plan.

The theory of specific cell structure also finds noteworthy support in the results of transplantation experiments; for heteroplasty has never proved successful, while tissues which are closely related will usually permit grafting. Pathology furnishes still further evidence in the fact that many poisons, such, for instance, as the tetanus toxin, will attack cells of a certain kind only—those of suitable structure—and leave all others uninjured.

The cell regulates its metabolism in finest detail, both qualitatively and quantitatively, and to accomplish this it must at all times enjoy unchanging conditions in the medium surrounding it. For a given location the composition of the plasma or lymph is always the same. Each cell takes from the plasma the same substances always, and gives to it always the same waste products, and though this fluid maintains for a given point a remarkable constancy of composition, it varies as it passes from place to place in the circulation.

Substances for which the body has use and which properly belong to the organism we call "body-like," while any substance which because of its structure and configuration fails to find adjustment in the organism is termed "body-foreign." Substances belonging to the plasma are "plasma-like," and those belonging peculiarly to the cell we call "cell-like" or "organ-like." Since the albumins of the plasma, as we have seen, must undergo reconstruction before they can become a part of the cell, though plasma-like they can only be regarded as cell-foreign. In like manner, materials belonging to a given cell while cell-like are plasma-foreign; for in the course of normal metabolism they find no opportunity for entering the plasma until through a step by step splitting at the hands of the cell they have been deprived of their specific character. A plasma-foreign substance entering the blood is equally foreign whether it come from the outside world or gain entrance to the plasma through some metabolic error on the part of the cell. The body must recognize it as foreign and deal with it accordingly, no matter what its source.

The animal organism should be regarded as a closed whole. Each organ or group of cells has its own specific duties to perform and contributes its definite part to the general metabolism. Disturb one and the whole machinery is upset. Introduce from the outside some substance which has no place in this general plan, or let some substance from within appear at a place not its own, and the organism feels the shock of foreign invasion.

Invasions by body-foreign for plasma-foreign substances are not uncommon. The bacterial infections and the malignant tumors serve as examples. Abderhalden has asked the question, "What course is open to the cells when body- or plasma-foreign substances gain entrance? Are they defenceless against these substances; or is it possible for the cells beyond the intestinal tract also to break down to harmless bodies such substances and use the building stones thus set free for their own purposes?"

In seeking an answer to this question, it was found that every body cell possesses at least some ferments similar in nature to those of the intestinal tract, and the conclusion is inevitable that these ferments play an all-important role in cell metabolism. It has been found that the body cells are able by means of a hydrolytic process to split fats into alcohol and fatty acids. Carbohydrates undergo similar degradation, glycogen being converted first into dextrin and then into maltose. This last-named is then split to dextrose. Numerous investigators have concerned themselves with inquiries regarding the presence of proteolytic or peptolytic ferments in the tissues. Abderhalden and the writer found no ferments in bovine plasma capable of splitting glycyl-l-tyrosin dl-alanyl-glycin, glycyl-dl-alanin, or diglycyl-glycin; while Abderhalden and Manwaring conducting parallel experiments with red and white cells from the same specimens of blood demonstrated the presence of such ferments. Investigators have almost without exception found peptolytic ferments in the other body cells. No such ferments are found in plasma alone, but there is no doubt that the body cells and the cells of the blood are equipped with ferments capable of digesting albumen.

The question is then asked, "Does the plasma defend itself, against foreign invasion by means of these normal cell ferments, or to meet foreign invasion does it take on new properties?" Experiment shows that the plasma on experiencing the shock of such invasion acquires new ferments,—ferments which are specific in action and are directed only against this particular plasma-foreign substance. If cane sugar be added to the serum obtained from a dog, no appreciable change appears within a reasonable time. If, however, the dog first receive intravenously a solution of cane sugar and after a short time the blood be drawn, it will be found that the serum so obtained has the power of converting cane sugar into dextrose. Invertin, the ferment which splits cane sugar, appears in the blood of the dog within fifteen minutes after a solution of this carbohydrate has been injected. Experimenting with the fat tributyrin, it was found that fats also stimulate the production of ferments specific for themselves.

The source of these defensive ferments is yet a matter of conjecture. No doubt all the organs, particularly the intestinal tract, the liver, and the kidneys, play a part, and the lively interest taken by the leukocytes in every form of infection or foreign invasion suggests for them an important role. On the other hand, recent observations indicate that defensive ferments are derived each from the organ against which its activities are directed.

If these facts be true, then it is possible for us to search for plasma-foreign substances by testing the blood-serum for their respective ferments. Upon this Abderhalden bases his test for pregnancy. The placenta, while body-like, is of course plasma-foreign. Experiment has shown that a living placenta gives off constantly, though perhaps in minute degree, its albumins to the mother circulation. The manner by which this plasma-foreign albumin gains entrance is as yet a matter of doubt. It has been suggested that the cells of the chorionic villi from time to time become dislodged and thus enter the maternal blood. In certain animals, however, the nature of the circulation precludes such an accident. Perhaps at this time of rapid cell activity a certain "overproduction" and "overflow" of a new albumin takes place. In whatever manner this invasion occurs, the mother within the first eight days of pregnancy exhibits in her serum a ferment which is capable of splitting placenta albumin and this only. The blood loses this ferment from fourteen to twenty-one days after the placenta is removed.

Were the practical application of these principles to stop here, Abderhalden's work with the defensive ferments would be of minor importance, since a blood-test for pregnancy is of no great general interest. The field is infinitely broader and richer. Invasion of the organism by body-foreign and plasma-foreign substances is an accompaniment not only of pregnancy but of many pathological states, and if the promise of this work is fulfilled, we shall be able to properly characterize these states—to recognize not only the plasma-foreign substance introduced from without, but also that introduced from within and the organ from which it comes. Through the serum diagnosis of pregnancy we are introduced to the much larger field of the serum diagnosis of organ function. We assume that when one organ becomes diseased or disturbed in function, the altered metabolism causes "unripe" or plasma-foreign albumins to enter the blood. We then test the blood-serum for the resulting specific ferments. It is tested separately against the albumins of all the organs in question, and the particular albumin digested indicates the organ which is diseased.

The opportunities thus opened for studies in physiology and pathology are almost unlimited. In this way the interrelation of the various organs may be studied: the mutual dependence of the organs of internal secretion, degenerations in muscle and nerve, bacterial infections, therapeutic and toxicologic questions, ad infinitum. "The whole realm of biology offers no more alluring task than the study of how the organism reacts when the harmony of its metabolism, regulated in finest detail, is disturbed."

Reports of work in this field and suggestions as to modification of technique are appearing in great profusion. The reports from Abderhalden's laboratory and from certain German clinics give wonderfully accurate results in pregnancy and in a variety of diseases. On the other hand, numerous investigators have shown disappointment in their results and express doubt as to the specific action of the ferments. The complexity of the method and the many pitfalls in the technique warrant always from its adherents the reply that the test was not properly carried out. All agree that before it comes into general use the present method must be changed or simplified, and Abderhalden has recently reported interesting and promising attempts at perfecting a precipitation method which will eliminate the dialysis shells. Certain of my own results have been contradictory, but one fruitful source of error has been discovered in the faulty dialysis shells. I hope to report these studies at another time.

Abderhalden stated recently that over 600 sera in his laboratory had been tested for pregnancy with less than 1 per cent. of error, and that these errors were later shown to have been avoidable. In the same report he tells of over 400 sera from cases of Graves' disease, dementia præcox, carcinoma, sarcoma, tuberculosis, and other diseases which have been tested with startlingly direct and accurate results. Fifty carcinoma sera were diagnosed without an error. In numerous instances serum diagnosis has pointed to certain diseased organs which could not have been recognized clinically.

Notwithstanding the many adverse criticisms, one cannot but recognize that such reports coming from men like Abderhalden, Lampé, and numerous others, cannot be disregarded.

Certain interesting results will serve to illustrate the value of the method. Sera from endemic goitre and myxedema are found to digest thyroid tissue. Exophthalmic goitre serum attacks thyroid, thymus, and frequently the ovary but no other tissue. Dementia præcox serum digests always the ovary or testis and usually the cerebral cortex. It has been reported that serum from patients with miliary tuberculosis will split the tubercle bacillus, and that serum from tuberculous cows splits bovine but not human tubercle bacilli.

Paltauf's case is worthy of mention. He found that a certain malignant tumor from an old woman was not split by known carcinoma serum, but was attacked by pregnancy serum. The *microscopic diagnosis proved to be malignant chorioepithelioma.*

Just a word regarding the present method. If the precautions prescribed are necessary (and the limited experience of the writer assures him that they are), no one can expect today to obtain satisfactory results who has not thoroughly familiarized himself first hand with Abderhalden's discussion of principles and his elaborate rules as to technique.

Briefly, the principle of the dialysis method of serum diagnosis, the method commonly used, rests upon the fact that colloidal albumin will not diffuse through an animal membrane, while its split products, peptones, and amino-acids, will pass through. The coagulated blood-free tissue of the placenta (or organ in question) is placed in a small dialysis shell, to which is added the serum to be tested. If this serum contains defensive ferments specific for this tissue, the latter will be digested and its split products will pass through to the distilled water outside the membrane—the dialysate—which after about sixteen hours can be tested with Ninhydrin for these bodies.[2]

Precautions are numerous and essential. Absolute asepsis in every step is demanded. The dialysis shells themselves must be standardized. They should not permit the passage of albumin, but must all be equally permeable to albumin-split products.[3] The preparation of the organ albumins, "substrata," as they are called, must be perfect. When properly prepared and coagulated they are snow-white. They must not contain even the minutest traces of blood or extractive substances. Before it is available for use, each preparation must be tested by the dialysis method for remaining traces of blood. Otherwise, results will be fallacious. All utensils must be in perfect condition both as to cleanliness and asepsis. The water used must be freshly distilled and sterile. Care must be taken to see that uneven evaporation of the different dialysates is impossible.

In applying the Ninhydrin test to the dialysates, the utmost precaution must be observed to obtain in each test identical intensity of boiling. To insure accurate observations as to the resulting color reaction, test-tubes of equal caliber and thickness of wall must be used. The blood-serum must be fresh, and should be actually, not apparently, free of cells. This work should not be done in a laboratory where other chemical or bacteriological experiments are carried out, and a special incubator should be reserved. The author of the method urges that those who undertake it familiarize themselves most carefully with its every detail, and begs that no attempt be made to change the method until we can first analyze our present errors and determine their sources.

And finally as to the part the clinician is to play in these studies. Abderhalden significantly states that physiology has done its part, that it has developed the method and its principles, and that it now remains for the clinician to determine value and scope. He emphasizes the necessity for the most careful clinical study and "follow up" of all cases tested by this method, and urges that patients be carefully classified and studied in large groups or series, rather than as individuals, as only the clinician is competent to do.

I take pleasure in acknowledging my indebtedness for the principles here discussed to the writings of Professor Abderhalden, and particularly to "Abwehrfermente des tierischen Organismus."

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[2] The optical method which is less adapted to clinical work depends upon the change in optical rotary power which a mixture of peptones undergoes when subjected to further splitting.

[3] The dialysis membranes now being brought on the market, because of the great demand and increased production, are almost universally unsuitable. The only reliable shells known to the writer are those which have been tested in Abderhalden's laboratory. They are sold by Rudolph Shoepps, Halle, a. s.